

# THE SPECIFICITY OF SEROLOGICAL REACTIONS

*Revised Edition*

BY

KARL LANDSTEINER, M.D.

*The Rockefeller Institute for Medical Research, New York*

With a Chapter on Molecular Structure and Intermolecular Forces by

LINUS PAULING



CAMBRIDGE, MASSACHUSETTS  
HARVARD UNIVERSITY PRESS

1945

## VIII

### MOLECULAR STRUCTURE AND INTERMOLECULAR FORCES

By LINUS PAULING

As our knowledge of the structure of molecules has greatly increased in recent years it has become clear that the physiological activity of substances is correlated not alone with their ability to take part in reactions in which strong chemical bonds are broken and formed, but also with the relatively weak forces which their molecules exert on other molecules. It is, indeed, probable that the high specificity which often characterizes physiological activity is in most cases specificity of intermolecular interaction rather than primarily of chemical reaction with the rupture and formation of strong bonds.

Atoms interact with other atoms in many ways. In the present discussion we divide interatomic forces into two classes, strong forces and weak forces. The strong forces are those which are responsible for the existence of stable molecules; they are the forces which lead to the formation of strong chemical bonds, with bond energies between 10 and 100 kilocalories per mole. The weak interatomic forces,

including van der Waals forces and "hydrogen-bond" forces, have energies of a few kilocalories per mole of interacting atom pairs; these forces are effective in holding molecules together without disrupting their individual structures, and also in operating between different parts of a large "loose-jointed" molecule in such a way as to hold it to a particular configuration.

The selection of the energy value of about 10 kilocalories per mole as the transition value between the two classes of interatomic interactions has its justification in statistical considerations based on thermal equilibrium at room temperature. Under ordinary circumstances at room temperature (that is, with the reacting substances present in reasonable concentrations) interaction of the "strong" class between two atoms will hold them together as a complex which is not significantly dissociated by the disrupting action of thermal agitation, whereas interaction of the "weak" class will not do this. Only by cooperation of the interactions between many atoms of one molecule and many atoms of another molecule can weak interatomic forces give rise to a stable intermolecular bond.

Weak interatomic forces have especial significance in serological phenomena—it is presumably these forces which lead to combination between an antibody molecule and a homologous antigen molecule or cell, and which also, operating between parts of the antibody molecule, are responsible for holding it in a configuration which confers on it the power of specific attraction for that antigen.

The interaction between molecules is determined in large part by the detailed structure of the molecules. In the following sections there are discussed first the nature of the chemical bond and the structure of molecules, and then the nature of intermolecular interactions.

#### THE STRUCTURE OF MOLECULES

The strong interatomic forces which lead to chemical bond formation are varied in nature. For convenience several extreme types, which are in general not sharply demarcated but show gradual transitions through intermediate types, are recognized; these are called ionic bonds, covalent bonds, metallic bonds, and other bonds of less importance.

Atoms such as sodium and chlorine can by losing or gaining electrons achieve stable electronic configurations, such as those of the noble gases. The resulting electrically charged ions with opposite

sign, such as  $\text{Na}^+$  and  $\text{Cl}^-$ , attract each other with the strong inverse-square Coulomb attraction. If the dielectric constant of the medium is small this attraction is very strong; it pulls the ions together until they are in contact. The size of an ion is determined by its electron distribution. Anions range in radius from 1.4 Å (for  $\text{F}^-$  and  $\text{O}^{2-}$ ) to 2.1 Å (for  $\text{I}^-$ ); and cations from very small values (about 0.6 Å for  $\text{Li}^+$ ,  $\text{Be}^{++}$ ,  $\text{Al}^{+++}$ ) to a maximum of 1.6 Å for  $\text{Cs}^+$ . When two ions are at the internuclear distance equal to the sum of their conventional crystal radii their outer electron shells interpenetrate to such an extent as to give rise to a repulsive force which balances the force of Coulomb attraction.

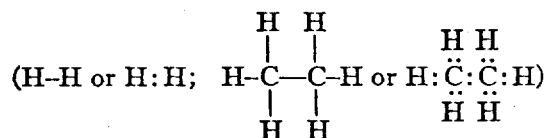
The energy of such an ionic bond in a medium of low dielectric constant is large—of the order of 100 kilocalories per mole—and the ionic bond is a strong bond. Strong ionic bonds exist between sodium ions and chloride ions in the sodium chloride crystal and in sodium chloride gas molecules (at high temperatures), and also, for example, between iron ions and magnesium ions and the surrounding nitrogen atoms of the porphyrin groups in hemoglobin and chlorophyll.

In water or other medium of high dielectric constant the Coulomb forces between ions are very greatly reduced in magnitude, so that in general they no longer lead to the formation of strong chemical bonds. The magnitude of the bond energy for Coulomb attraction of two electrically charged atoms or groups can be easily calculated; it is the product of the two electrical charges divided by the distance between them and by the dielectric constant of the medium. For example, the Coulomb bond strength for a carboxyl ion group  $\text{RCOO}^-$  and an ammonium ion group  $\text{RNH}_3^+$ , which can approach each other until the center of the nitrogen atom comes to within about 3 Å of the center of an oxygen atom, is calculated to be about 4.5 kcal. per mole for a medium with dielectric constant 25 (the effective value for water for two charges at this distance<sup>1</sup>). This bond energy is not great enough to cause the two groups to form a stable aggregate; however, as is mentioned in a later section, hydrogen-bond formation between the groups increases the strength of their interaction greatly, and together with the direct ionic attraction gives rise to a rather strong bond between the groups.

Of by far the greatest importance in determining the structure of organic molecules is the covalent bond or shared-electron-pair bond. This is the ordinary valence bond of the organic chemist. The single

<sup>1</sup> Schwarzenbach (1).

covalent bond results from the sharing of an electron pair (two electrons) between two atoms

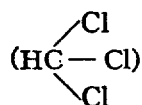


the double bond from the sharing of two electron pairs ( $\text{H}_2\text{C}=\text{CH}_2$  or  $\text{H}_2\text{C}::\text{CH}_2$ ), and the triple bond from the sharing of three electron pairs ( $\text{HC}\equiv\text{CH}$  or  $\text{HC}:::\text{CH}$ ). The carbon atom usually forms four covalent bonds (counting the double bond as two and the triple

bond as three), the nitrogen atom three (as in the amines,  $\begin{array}{c} \text{R} \\ | \\ \text{:N}-\text{R} \\ | \\ \text{R} \end{array}$ )

or four, in which case it assumes a positive charge  $\begin{array}{c} \text{H} \\ | \\ (-\text{CH}_2-\text{N}^+-\text{H}) \\ | \\ \text{H} \end{array}$

the oxygen atom two ( $(\text{CH}_3)_2\text{C}=\text{O}$  or  $\text{CH}_3-\text{O}-\text{CH}_3$ ) or one ( $-\text{O}-\text{H}$ ), and halogen atoms one



or zero (chloride ion,  $\text{Cl}^-$ ).

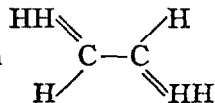
The covalent bond holds atoms compactly together: the C—C single bond distance is 1.54 Å, the C=C double-bond distance 1.33 Å, etc. The values of these and many other interatomic distances have been found by experimental investigations, mainly by the diffraction of x-rays and electrons, and it is now possible to predict with confidence, in most cases to within 1 or 2 %, the interatomic distances for any molecule of known chemical structure.

The remaining information needed to define the configuration of a covalently bonded molecule relates to the mutual orientation in space of the bonds. For the atoms which in the main occur in molecules of biological substances single bonds usually are directed toward the corners of a regular tetrahedron, making with one another approximately the tetrahedral angle  $109^\circ 28'$ . The picture of the tetrahedral angle of Van't Hoff and Le Bel similarly leads to the

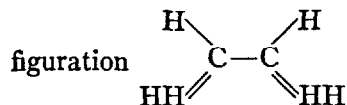
following values of angles involving multiple bonds: double bond and single bond,  $125^{\circ}16'$ ; two double bonds,  $180^{\circ}$ ; triple bond and single bond,  $180^{\circ}$ . These predictions have all been verified as holding to within a few degrees by extensive structural studies (electron diffraction, x-ray diffraction) carried out in recent years.

The rotational configuration about a double bond is highly restricted by the requirement that the system, including the adjacent single bonds, be coplanar. Except for possible uncertainty in identification of *cis* and *trans* configurations, this requirement specifies uniquely the configuration about the double bond.

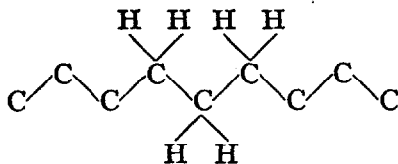
The orientation about a single bond is less well defined. It has been discovered recently that the "staggered" orientation



for ethane and related molecules is more stable by a small amount of energy (about 3 kilocalories per mole) than the "eclipsed" con-



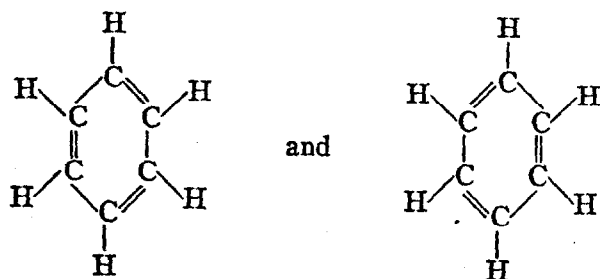
which differs from the "staggered" configuration by rotation of one group through the angle  $60^{\circ}$  relative to the other. However, the requirement that each single bond have the "staggered" orientation often leaves accessible several alternative configurations for a complex molecule among which the molecule easily changes. For a hydrocarbon chain, for example, there exist alternatives such as the extended configuration



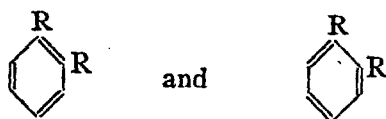
and the various coiled configurations which result from this by rotating through  $120^{\circ}$  about any of the carbon-carbon bonds. X-ray and electron-diffraction studies have shown that molecules of the normal hydrocarbons and related substances have the extended configuration in crystals, but also assume other coiled configurations in the vapor state at higher temperatures.

The phenomenon of *resonance among alternative valence-bond*

*structures* is of great importance in determining the configuration of aromatic and conjugated systems. For example, for the benzene molecule the two reasonable valence-bond structures



(the Kekulé structures) can be written. These structures, although they are the most satisfactory single structures proposed for the molecule, are unsatisfactory in several ways. Thus benzene does not show the unsaturation which would be expected for a molecule containing double bonds, and it does not form the pairs of isomeric compounds, such as

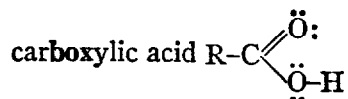


which would be expected if the individual Kekulé structures existed independently. The solution of these and many other difficulties has been provided recently by the idea, which is a consequence of the physical theory of quantum mechanics, that a molecule such as benzene, for which two or more alternative reasonable electronic structures can be written, is represented not by any one of these structures but by a combination of them; the molecule is described as resonating among all of the alternative structures, and its properties are determined by all of them. For benzene, which resonates between the two Kekulé structures, each of the carbon-carbon bonds has 50% double-bond character; this gives each carbon-carbon bond the stereochemical properties (coplanarity) of a double bond, which requires the molecule to be a completely coplanar regular hexagon and to have carbon-carbon bond distances (1.39 Å) between the single-bond and double-bond values.

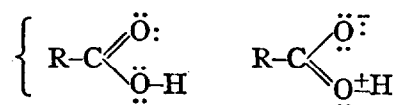
A very important consequence of the theory of resonance is that

a molecule which resonates between two or more alternative electronic structures is more stable thermodynamically than it would be if it had any one of the structures alone. This energy of stabilization, called the *resonance energy*, amounts for benzene to 39 kcal. per mole. The benzene molecule is more stable by 39 kcal. per mole than it would be if it had a non-resonating Kekulé structure; and since similar resonance stabilization does not occur for its addition compounds, benzene is much more resistant to hydrogenation and similar reactions than are molecules containing non-resonating double bonds. In the same way the resonance energy of other aromatic molecules stabilizes them and causes them to behave as saturated rather than unsaturated substances.

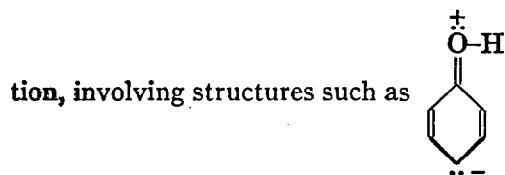
It might be expected that the OH group in acetic acid or other



would dissociate to about the same extent as in the alcohols. Experiment shows, however, that the dissociation constants for these acids are very much larger (by factors of about  $10^{12}$ ) than those for the alcohols. The explanation of this fact provided by the theory of resonance is that the carboxylic acids have the resonating structure

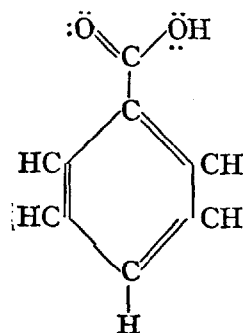


and the contribution of the second of these structures, which assigns a carbon double bond to the oxygen atom with hydrogen attached, causes the proton to tend to leave the molecule. A similar explanation,

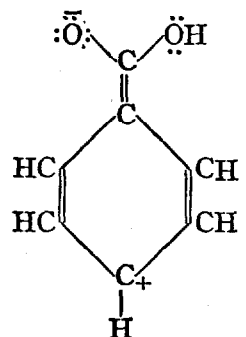


as well as the normal structures, accounts for the greater acidity of phenols as compared with aliphatic alcohols. In a conjugated system such as benzoic acid





in which the double bond of the carboxyl group is conjugated with the double bonds of the benzene ring, the connecting single bond has enough double-bond character to keep the carboxyl group coplanar with the ring. This double-bond character results from resonance with structures such as



Steric interactions between atoms in a molecule are often important in preventing the assumption of an otherwise stable configuration. Thus the carboxyl group in *o*-methylbenzoic acid is prevented from becoming coplanar with the benzene ring by the steric repulsion between the methyl group and the carboxyl oxygen atoms.

The examples given above do no more than to indicate the breadth of the field of application of the theory of resonance. Many further applications are discussed in treatises on molecular structure and theoretical organic chemistry.<sup>2</sup>

<sup>2</sup> Pauling (2), Hammett (3), Branch et al. (4), Rice (5), Remick (13).

## INTERMOLECULAR INTERACTIONS

Two molecules may attract each other through interactions classified as electronic van der Waals attraction, Coulomb attraction of groups with opposite electric charges, attraction of electric dipoles or multipoles, hydrogen-bond formation, etc. Whatever the nature of the attraction may be, the forces of attraction increase in general as the molecules approach one another more and more closely, and the bond between the molecules reaches its maximum strength when the molecules are as close together as they can come. The molecular property which determines the distance of closest approach of two molecules is the electronic spatial extension of the atoms in the molecules. It is possible to assign to each atom a *van der Waals radius*, which describes its effective size with respect to intermolecular interactions. These radii vary in value from 1.2 Å for hydrogen through 1.4–1.6 Å for light atoms (fluorine, oxygen, nitrogen, carbon) to 1.8–2.2 Å for heavy atoms (chlorine, sulfur, bromine, iodine, etc.). The shape of a molecule can be predicted by locating the atoms within the molecule with use of bond distances and bond angles and then circumscribing about each atom a spherical surface corresponding to its van der Waals radius. This shape determines the ways in which the molecule can be packed together with other molecules.

The most general force of intermolecular attraction, which operates between every pair of molecules, is *electronic van der Waals attraction*. This type of electronic interaction between molecules was first recognized by London (6). A molecule (of argon or carbon tetrachloride, for example) which has no permanent average electric dipole moment may have an instantaneous electric dipole moment, as the center of charge of the electrons, in their rapid motion in the molecule, swings to one side or the other of the center of charge of the nuclei. This instantaneous dipole moment would produce an instantaneous electric field. Any other molecule in the neighborhood would be polarized by this field, which would cause its electrons to move relative to its nuclei in such a way as to give rise to a force of attraction of the second molecule toward the first. By the time that this electronic polarization had occurred the electrons of the first molecule would have moved some distance, and its instantaneous dipole moment would have changed in value. It is clear that the theoretical discussion of the phenomenon is not simple; it was carried out by London, who found that the effect as calculated by quan-

tum-mechanical methods corresponds closely in magnitude with the observed van der Waals attraction for various simple substances (noble gases, hydrogen, oxygen, methane, etc.), and explains quantitatively the intermolecular attraction which leads to their condensation to form liquids.

This electronic van der Waals attraction operates between every atom in a molecule and every atom in other molecules in the near neighborhood. The force is stronger for heavy atoms than for light atoms. It increases very rapidly with decreasing interatomic distance, being inversely proportional to the seventh power of the interatomic distance. Because of this, the electronic van der Waals attraction between two molecules in contact is due practically entirely to interactions of pairs of atoms (in the two molecules) which are themselves in contact; and the magnitude of the attraction is determined by the number of pairs of atoms which can be brought into contact. In consequence, two molecules which can bring large portions of their surfaces into close-fitting juxtaposition will in general show much stronger mutual attraction than molecules with less extensive complementarity of surface topology.

A measure of the energy of the van der Waals attraction of molecules is given by the heats of sublimation of their crystals. This energy is small for light molecules and larger for heavier molecules. For the hydrogen molecule it is about 0.25 kcal. per mole, for nitrogen and oxygen molecules about 1.9, for argon 2.0, for krypton and methane 2.7, and for xenon 3.8. For large molecules the energy of van der Waals attraction is correspondingly larger, in proportion to the number of atoms in the molecule.

Except for molecules with very large electric dipole moment or capable of forming hydrogen bonds, the van der Waals attraction is responsible for the major part of the interaction with other molecules.

Other types of molecular interactions result from the possession by one or both of the interacting molecules of a permanent electric dipole moment or electric moment of higher order. The effects of these permanent moments have been classified in various ways, as dipole-dipole forces, the forces of electronic polarization of one molecule in the dipole field of another, etc. In general, however, these electric forces are of minor importance, except when an isolated or essentially isolated electric charge is involved. Thus for hydrogen chloride, with a rather large value of the electric dipole moment, about five-sixths of the energy of intermolecular attraction is due

to electronic van der Waals attraction and only about one-sixth is the result of dipole interaction.

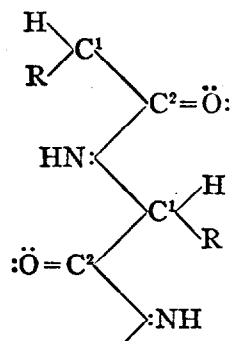
A type of intermolecular attractive force which ranks in importance with the electronic van der Waals attraction is that associated with the structural feature called the hydrogen bond. The importance and generality of occurrence of the hydrogen bond were first pointed out in 1920 by Latimer and Rodebush (7). A hydrogen bond results from the attraction of a hydrogen atom attached to one electronegative atom for an unshared electron pair of another electronegative atom. The strength of a hydrogen bond depends on the electronegativity of the two atoms which are bonded together by hydrogen; fluorine, oxygen, and nitrogen, the most electronegative of all atoms, are the atoms which form the strongest hydrogen bonds. The energy of a hydrogen bond between two of these atoms is of the order of magnitude of 5 kcal. per mole. This is so large as to have a very important effect on the intermolecular interactions of molecules capable of forming hydrogen bonds and on the properties of the substances consisting of these molecules. It is hydrogen-bond formation between water molecules which gives to water its unusual physical properties — abnormally high melting point, boiling point, heat of fusion, heat of vaporization, dielectric constant, etc. Hydrogen-bond formation is also responsible for the existence of the hydrogen fluoride ion  $\text{HF}_2^-$ , and for the polymerization of hydrogen fluoride, the carboxylic acids, and other substances.

The hydrogen bond is not specific, inasmuch as a hydrogen atom of a hydroxyl group, for example, will attract the unshared electron pair of any electronegative atom which comes into its neighborhood. But the attracted atom must be able to come to a definite position in space in order that a stable hydrogen bond may be formed. This position is along the line of the OH axis (for a hydroxyl group) and at a determined distance, which is about 2.7 Å for an O-H-O hydrogen bond. This steric restriction and the limitation of the class of atoms capable of forming good hydrogen bonds give to the hydrogen bond somewhat greater stereochemical significance than is shown by the electronic van der Waals attraction.

The third type of attractive force which is of significance in protein structure is the interaction of electrically charged groups, such as the carboxyl ion side chains and ammonium ion side chains of amino acid residues. As stated above, the direct electrostatic interaction of two such groups amounts to about 4.5 kcal. per mole. However, the hy-

drogen atoms of the ammonium ion group are able to form hydrogen bonds with the oxygen atoms of the carboxyl ion group, increasing very much the energy of the interaction of these two groups, and permitting a stable complex, with bond energy of the order of 10 kcal. per mole, to be formed.

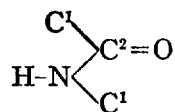
The peptide chain provides illustration of several structural features.



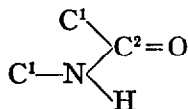
The conventional electronic structure shown above does not provide a complete representation of the chain; each peptide group shows resonance of the type



which gives the C<sup>2</sup>-N bond some double-bond character. In consequence of this and the stereochemical property of coplanarity about double bonds, the stable configurations of the chain are those which make coplanar the group of atoms

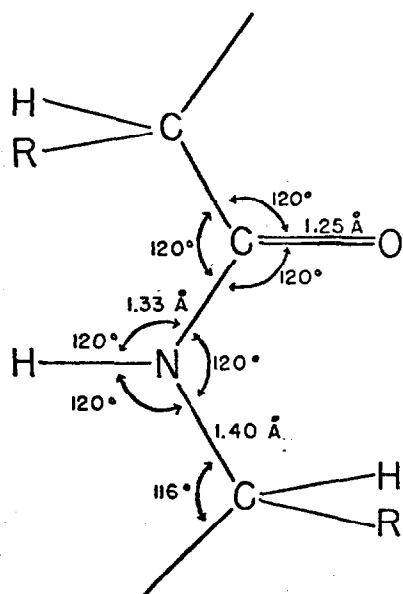


There are two such configurations; the extended one given above, and the contracted one

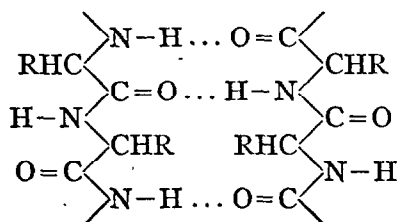


In addition, the chain has considerable freedom of orientation about

the single bonds C<sup>1</sup>-C<sup>2</sup> and N-C<sup>1</sup>. Further selection among the many corresponding configurations for the peptide chain results from the requirement that no two non-bonded atoms of the chain can come closer together than their limiting contact distance, the sum of their van der Waals radii. Interatomic distances and bond angles for the fully extended peptide chain, as given by Corey (8), are

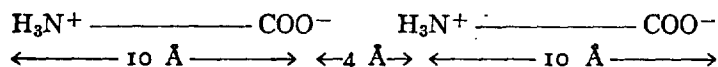


Another result of the resonance of the double bond in the peptide group is that the oxygen atom receives an increased negative charge and the nitrogen atom an increased positive charge. Both of these effects tend to increase the tendency of these atoms to take part in hydrogen-bond formation, and in consequence we would expect stable configurations of protein molecules to be those in which many hydrogen bonds between peptide chains are formed. Such a structure, for extended polypeptide chains, is the following:



A structure of this type was first suggested, for  $\beta$ -keratin, by Astbury, on the basis of his x-ray studies.

Whereas a clear distinction can be made between electronic van der Waals forces and other intermolecular forces, designated as Coulomb attraction of ions, attraction of electric dipoles and multipoles, and hydrogen bonds, these latter types are not clearly demarcated. A neutral molecule containing a carboxyl-ion group  $\text{COO}^-$  and an ammonium-ion group  $\text{NH}_3^+$  may be described as having an electric dipole moment, and the force of attraction between two such zwitterionic molecules may be said to result from the interaction of two electric dipoles; this, however, is not the best way of describing the system except when the two molecules are very far apart. If the molecules are close together, the carboxyl-ion group of one molecule may be very near to the ammonium-ion group of the other — within 4 Å — with the other charged groups much farther away, perhaps



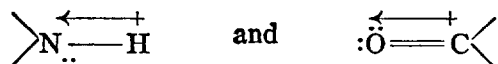
10 Å or 20 Å away if the molecules are large. The intermolecular attraction for this configuration will be very nearly the same as for a carboxyl ion and an ammonium ion at 4 Å; and it can be expressed still more closely as the sum of the Coulombic interactions of the charged groups in pairs.

Similarly the forces between two protein molecules may be conveniently described in terms of the total electric charges on the molecules and their electric dipole moments and multipole moments so long as the molecules are far apart (compared with their own diameters of say 50 or 100 Å); when the molecules approach one another more closely, and especially when they come into contact, the forces are more conveniently discussed by considering the interactions of small parts of one molecule and small parts of the other.

Since the distances between atoms in contact are about 3 Å or 4 Å, and the dimensions of usually recognized atomic groups are not much greater, electric charges in a molecule which are more than about 5 Å from other charges in the molecule may conveniently be considered separately, whereas pairs of opposite charges less than 5 Å apart in the same group may be described as forming the electric dipole of the group. The dipole moments of groups are usually small — less than  $2 \times 10^{-18}$  e.s.u. This moment corresponds to unit positive and negative charges (equal in magnitude to the electronic charge) sep-

arated by less than 0.5 Å. It is seen that the force of attraction of two such dipoles or of a dipole and an ion at a distance of 4 Å or more would be very much less than the Coulomb attraction of two ionic groups, since the effect of a positive charge would be nearly completely neutralized by the opposite effect of the nearby negative charge. It is for this reason that the dipole and multipole moments of groups make little contribution to the forces between molecules, compared with ionic groups. Moreover, as mentioned above for hydrogen chloride, the intermolecular forces due to dipole moments are also much smaller than those due to electronic van der Waals attraction.

The hydrogen bond may be said to arise in large part from the attraction of the dipole moments of the groups involved, such as



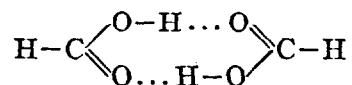
(the arrow pointing from the positive charge toward the negative charge of the dipole), with the dipole-dipole attraction unusually large because of the close approximation of the groups permitted by the small size of the hydrogen atom. Other factors (specific interaction of the proton and an electron pair of the electronegative atom of the approaching group) also play a part, however, and it is customary to designate this special sort of interaction of groups as hydrogen-bond formation.

The theoretical discussion of the interaction of molecules in solution is complicated by competition with solvent molecules. A solute molecule may have as strong electronic van der Waals attraction for the solvent molecules as for other solute molecules. The electronic van der Waals attraction of atoms for all other atoms operates in every condensed system to bring all the atoms into as close packing as is permitted by the chemical bonds which determine the shapes of the molecules and by the other intermolecular forces. This is the reason that the molal volumes of most liquids and crystals can be expressed to within a few percent as the sums of definite atomic volumes. Only rarely (as in the case of ice, in which a rather open structure is stabilized by hydrogen-bond formation at tetrahedral angles) do exceptions occur. The effective electronic van der Waals attraction of two solute molecules in aqueous solution may be very small, since the close approach of the two molecules involves the replacement of the water molecules adjacent to each molecule. Sim-



ilarly the effective hydrogen-bond attraction between two solute molecules is large only if the solute-solute hydrogen bonds plus the corresponding water-water hydrogen bonds are significantly stronger than the ruptured solute-water hydrogen bonds.

Of the great number of well-defined intermolecular compounds which are known, precise structural information is available for only a few; it is only very recently that x-ray and electron-diffraction methods and related techniques have been sufficiently developed to permit them to be applied to complexes containing more than a dozen atoms or so, and their extensive successful application in this field has not yet been accomplished. Reliable information has been obtained about the dimer of formic acid



and about a number of other polymers in which the intermolecular attraction is due to hydrogen bonds. Detailed studies have also been made of Werner coordination complexes, such as the hydrated and ammoniated ions  $\text{Zn}(\text{NH}_3)_4^{++}$ ,  $\text{Ni}(\text{H}_2\text{O})_6^{++}$ ,  $\text{Co}(\text{NH}_3)_6^{+++}$ , etc. The forces which hold the water or ammonia molecules to the central ion are in part the electrostatic attraction of the ion for the electric dipole molecules; more important is the formation of covalent bonds between the central atom and the attached molecules, with use for each bond of an electron pair provided by the molecule. Most atoms tend to form four bonds at tetrahedral angles (or in special cases four coplanar bonds directed toward the corners of a square) or six bonds directed towards the corners of a regular octahedron. The formation of bonds of this sort may be of general importance in holding the metal-containing prosthetic groups to their protein molecules in the respiratory pigments and related substances. There is strong evidence indicating that in hemoglobin each protoheme is held to globin not only by Coulomb attraction between the propionate ion side chains and positively charged groups in the globin but also by the formation of an octahedral bond between the iron atom in the center of the four nitrogen atoms of the porphyrin and a nitrogen atom of the imidazole ring of a histidine residue in the globin. It seems likely that in some heme pigments, including cytochrome c, each iron atom forms two bonds in this way with two parts of the protein molecule. Similar bonds are probably present also in hemocyanin and other

pigments containing metal atoms, aiding in holding the complexes together.

Because of the lack of detailed structural information, there is doubt as to the nature of the forces which lead to compound formation between nitro compounds such as picric acid and various aromatic hydrocarbons. It is probable that the electric dipoles of the nitro groups are important here, and that the high polarizability of aromatic molecules in the plane of the rings gives rise to strong attraction to the nitro molecule when it is suitably oriented.<sup>3</sup> The unusually strong bonds formed between antibody molecules and some nitro haptens<sup>4</sup> may result from a similar effect.

An extraordinary set of molecular compounds is formed by certain sterids, especially desoxycholic acid. In the compounds between desoxycholic acid and various carboxylic acids, dicarboxylic acids, esters, and other molecules it is found<sup>5</sup> that one molecule of the latter combines with 1, 2, 3, 4, 6, or 8 molecules of desoxycholic acid, the number increasing with increase in size of the molecule (1 for acetic acid, 2 (or 3) for propionic acid, and so on to 8 for pentadecylic and larger acids). That hydrogen bonds are involved in the formation of these complexes is indicated by the fact that desoxycholic acid and also  $\alpha$ -apocholic acid and  $\beta$ -apocholic acid, which form similar complexes, contain two hydroxyl groups, in essentially the same stereochemical configuration, whereas other related substances without hydroxyl groups or with different configurations are ineffective. It is likely that several molecules of desoxycholic acid pile up in such a way that a cavity is formed through them into which fits the foreign molecule, which thus serves to key them together; but experimental substantiation of this surmise is lacking.

The forces of van der Waals attraction, hydrogen-bond formation, and interaction of electrically charged groups are in themselves not specific; each atom of a molecule attracts every other atom of another molecule by van der Waals attraction, each hydrogen atom attached to an electronegative atom attracts every other electronegative atom with an unshared electron pair which comes near it, and each electrically charged group attracts every other oppositely charged group in its neighborhood. Similarly the van der Waals repulsive forces are non-specific; each atom in a molecule repels

<sup>3</sup> Briegleb (9), Pauling (10).

<sup>4</sup> Pressman, Brown and Pauling (11).

<sup>5</sup> For references see Sobotka (12).

every other atom of another molecule, holding it at a distance corresponding to the sum of the pertinent van der Waals radii.

We see, however, that specificity can arise in the interaction of large molecules as a result of the spatial configuration of the molecules. Two large molecules may have such shapes that the surface of one cannot be brought into contact with the surface of the other except at a few isolated points. In such a case the total electronic van der Waals attraction between the two molecules would be small, because only the pairs of atoms near these few isolated points of contact would contribute appreciably to this interaction. Moreover, the distribution of positively and negatively charged groups and of hydrogen-bond forming groups of the two molecules might be such that only a small fraction of these groups could be brought into effective interaction with one another for any position and orientation of one molecule with respect to the other. The energy of attraction of these molecules would then be small. If, on the other hand, the two molecules possessed such mutually complementary configurations that the surface of one conformed closely to the surface of the other, there would be strong electronic van der Waals attraction between all of the atoms on the surface of one of the molecules and the juxtaposed atoms of the complementary surface of the other molecule. And if, moreover, the electrically charged groups of one molecule and those of the other were so located that oppositely charged groups were brought close together as the molecules came into conformation with one another, and if the hydrogen-bond forming groups were also so placed as to form the maximum number of hydrogen bonds, the total energy of interaction would be very great, and the two molecules would attract one another very strongly. It is clear that this strong attraction might be highly specific in the case of large molecules which could bring large areas of their surfaces into contiguity. A molecule would show strong attraction for that molecule which possessed completely complementariness in surface configuration and distribution of active electrically charged and hydrogen-bond forming groups, somewhat weaker attraction for those molecules with approximate but not complete complementariness to it, and only very weak attraction for all other molecules.

This specificity through complementariness of structure of the two interacting molecules would be more or less complete, depending on the greater or smaller surface area of the two molecules involved in the interaction. It may be emphasized that this explanation of spe-

cificity, as due to a complementariness in structure which permits non-specific intermolecular forces to come into fuller operation than would be possible for non-complementary structures, is the only explanation which the present knowledge of molecular structure and intermolecular forces provides.

## BIBLIOGRAPHY

- (1) Schwarzenbach: *ZP A* 176, 133, 1936. — (2) Pauling: *The Nature of the Chemical Bond*, Cornell University Press, Ithaca, 2d ed., 1940. — (3) Hammett: *Physical Organic Chemistry*, McGraw-Hill Book Co., New York, 1940. — (4) Branch et al: *Theory of Organic Chemistry*, Prentice Hall, New York, 1941. — (5) Rice: *Electronic Structure and Chemical Binding*, McGraw-Hill Book Co., New York, 1940. — (6) London: *Z. f. Physik* 63, 245, 1930. — (7) Latimer et al: *JAC* 42, 1419, 1920. — (8) Corey: *ChR* 26, 227, 1940. — (9) Briegleb: *ZP B* 26, 63, 1934, *B* 31, 58, 1935. — (10) Pauling: *PNA* 25, 577, 1939. — (11) Pressman et al: *JAC* 64, 3015, 1942. — (12) Sobotka: *The Chemistry of the Steroids*, Williams and Wilkins Co., Baltimore, 1938. — (13) Remick: *Electronic Interpretations of Organic Chemistry*, John Wiley and Sons, Inc., New York, 1943.